EDITORIAL

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Polymerase Chain Reaction Surveillance of Microbial DNA in Critically Ill Patients: Exploring Another New Frontier

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Surgeons continue to be plagued by infectious complications in their patients, and appropriate and timely diagnosis and treatment remain central to reducing this morbidity and mortality. Precise bacteriologic detection of the invading microorganisms is essential to contemporary surgical care, and this is commonly accomplished by culturing wound, cavitary drainage, other body fluids and blood. Such tests are time-consuming and costly; conclusive results are rarely available until 48 hours after samples were obtained. Even more frustrating are the circumstances that surround the bacteriologic report of "no growth" on blood cultures taken from a critically ill patient deemed to be septic. A recent consensus review of the new biology as it relates to critical care suggests that these patients have the systemic inflammatory response syndrome (SIRS)[1]; bacteremias are found in 30% to 50% or so of patients with this diagnosis. Therapy remains ill defined for most of the patients with inflammation but without infection.

The discovery that signal proteins elaborated from our bodies' own cells account for much of the cardiovascular, metabolic, and immunologic response to infection and inflammation has caused a re-evaluation of the role played by the infectious agent *versus* the host's response.[2] A new paradigm is emerging that suggests that we should be more concerned with the host's responses than the infectious organism. As a result, some have suggested that we monitor cytokine levels in the blood and relate the presence of these and other substances to physiologic or immunologic events. If particular patterns emerged, these could be related to specific infections and highly directed antibody and antimicrobial therapy could be initiated.

Such a search, however, has failed to reveal a motherlode of diagnostic information. We have learned that levels of interleukin-1β are often elevated in septic patients,[3] that levels of tumor necrosis factor are more commonly elevated at the time of bacteremias,[4] and that levels of interleukin-6 (IL-6) are almost always elevated. Like serum C-reactive protein, IL-6 elevation is a reasonable indicator of the extent of the inflammatory process[5]: the higher the level, the greater the inflammation. However, this chapter is still being written, because more cytokines and other inflammatory factors (*e.g.*, endotoxins, leukotrienes, complement) are being discovered and quantified. The pharmacologic approach continues in an attempt to block or bind these substances and attenuate their responses. This area of research may well evolve into a new diagnostic computer-generated form of pattern recognition in the years to come. Such information ideally could help in the treatment of severely ill patients with sepsis or SIRS.

In this issue of *Annals of Surgery*, investigators report an entirely new approach to the diagnostic dilemma posed by the patient with sepsis or SIRS: the detection of bacterial DNA in the blood of critically ill patients.[6] Wesley Alexander et al.[6] have long sought to understand the various factors that affect host resistance and cause the surgical patient to become susceptible to infectious complications. These investigators used a discovery that is a product of the revolution in cell biology. Scientists have needed to determine the composition of small fragments of DNA to enhance understanding of the specific elements of genetic coding. Investigators took advantage of the ability of DNA to replicate itself: using the polymerase chain reaction (PCR) technique, DNA fragments, when incorporated with appropriate primers and exposed to optimal conditions, can be copied, and when sufficient sample is available, the specimen can be analyzed. Using

these techniques, Kane et al.[6] examined blood samples from controls, ICU patients, and organ transplant patients who had received the monoclonal antibody OKT3. Blood samples from these patients were screened for three different types of bacterial DNA: one derived from the B-glucosidase gene of *Escherichia coli,* another from the ever-present glutamine synthase gene found in many *Bacteroides* species, and a third from a highly conserved area of DNA found in 12 common species of both gram-negative and -positive organisms.

Previous studies in the investigators' laboratory determined the sensitivity of the technique to be on the order of 100 fg of template DNA, a quantity thought to represent only 10 microorganisms![7] Sensitivity is great, but specificity is a problem: if DNA is detected, it is unknown if it represents live invading organisms or dead presorbed organisms. Alternatively, the DNA could come from organisms that have been engulfed and killed by phagocytes.

Given these provisos, and recognizing that the DNA screen may not detect all bacterial DNA present in the blood, the authors correlated the presence of bacterial DNA with the patient's clinical course and the blood culture data. Three ICU patients became bacteremic, and all three were PCR-positive for bacterial DNA. Other ICU patients had established infections with positive cultures obtained from wound, lung, peritoneal fluid, and so forth; all these patients had bacterial DNA in their blood, although their blood cultures were negative. Additional surgical patients studied had no sites of infection, but 36% of the patients in this group were PCR-positive. It is unknown whether this represents a false-positive rate or whether some ICU patients without established infection naturally have bacterial DNA in their blood.

An intriguing subplot of this study is the response of a group of transplant patients who exhibited chills, fever, and other systemic signs of sepsis after OKT3 administration. This response, dubbed the cytokine release syndrome, was associated with the presence of bacterial DNA in the blood of all eight patients, although only one of the eight had a positive blood culture. Could the OKT3 enhance bacterial translocation from the intestinal tract or elsewhere to cause this symptom complex?

This intriguing work from a laboratory that has long led the effort to understand surgical infections has provided us with a provocative report with many unanswered questions. However, this is the nature of many initial steps in the discovery process. More information is now required to answer these and other questions: How is bacterial DNA cleared from the body? Is the presence of bacterial DNA in the blood the result of increased bacterial invasion or enhanced release of these substances from phagocytic cells? How does the presence of bacterial DNA in the blood relate to the clinical signs and symptoms of sepsis (*e.g.*, time courses and dose-response data)? If we infused these DNA components into animals or volunteers, would we reproduce the sepsis syndrome? Will detection of these molecules enhance our understanding of surgical infections? Will this measurement system somehow enhance clinical care?

Drs. Kane, Alexander, and Johannigman have taken a leadership role by applying new technology to help understand and solve an age-old surgical problem, that of surgical infection. This report should stimulate others to use this methodology to understand better the pathophysiology of infections and inflammation in critical illness and to define the relations between invasive microorganisms and the host over exuberant inflammatory response. Only by understanding more about infection and the multifaceted aspects of the host's response will we be able to improve our therapies for this costly complication and provide better care for our patients.

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